Clin Chem Lab Med 2009;47(8):1001–1004 © 2009 by Walter de Gruyter • Berlin • New York. DOI 10.1515/CCLM.2009.216

Unstimulated high sensitive thyroglobulin measurement predicts outcome of differentiated thyroid carcinoma

Luca Giovanella[1,2,*], Marco Maffioli[3], Luca Ceriani[1], Diego De Palma[4] and Giuseppe Spriano[5]

[1] Department of Nuclear Medicine, Oncology Institute of Southern Switzerland, Bellinzona, Switzerland
[2] Department of Clinical Chemistry, Oncology Institute of Southern Switzerland, Bellinzona, Switzerland
[3] Department of Head and Neck Surgery, University Hospital ''Fondazione Macchi'', Varese, Italy
[4] Department of Nuclear Medicine, University Hospital ''Fondazione Macchi'', Varese, Italy
[5] Department of Head and Neck Surgery, Oncology Institute ''Regina Elena'', Rome, Italy

Abstract

Background: Thyroglobulin (Tg) measurement following thyrotropin (TSH) stimulation is used in the follow-up of patients with differentiated thyroid carcinoma (DTC). However, high-sensitive assays allow accurate measurement of serum Tg even without TSH stimulation. Here, we prospectively evaluated the impact of unstimulated high-sensitive Tg measurement in early and long-term outcome of patients with DTC.
Methods: One hundred and ninety five patients affected with DTC were evaluated. Six months after thyroid ablation (i.e., thyroidectomy plus radioiodine) serum Tg was measured during TSH-suppressive thyroxine (T4) treatment (onT4-Tg). Patients with undetectable onT4-Tg and negative neck ultrasound (US) were considered disease free and onT4-Tg was measured every 12 months for a mean follow-up of 6.8 (4.7–8.9) years. Patients with an increase in onT4-Tg underwent specific diagnostic work-up and appropriate treatment if necessary.
Results: Four patients showed recurrence at first follow-up visit with a corresponding increase in onT4-Tg concentrations (sensitivity 100%). Three patients had false positive onT4-Tg measurement (specificity 98%) with a spontaneous decrease within 3–6 months in all cases (specificity 100%). Three of 188 patients with undetectable serum onT4-Tg at first follow-up showed recurrence later with an increase in onT4-Tg as the first (n=2) or unique (n=1) sign of relapse (sensitivity 100%). Among 185 disease-free patients in a prolonged follow-up, 12 had a transient increase in onT4-Tg (specificity 91.6%). However, a spontaneous reduction within 3–6 months occurred in all cases (specificity 100%).
Conclusions: Undetectable serum onT4-Tg using a high-sensitivity immunoradiometric assay 6 months after thyroid ablation predicts low-risk of DTC recurrence. When onT4-Tg became detectable during follow-up, the evaluation of Tg slope in a 3–6 months period accurately discriminated patients with DTC recurrence from those without recurrence. This helped avoid unnecessary diagnostic or therapeutic procedures.
Clin Chem Lab Med 2009;47:1001–4.

Keywords: differentiated thyroid carcinoma (DTC); follow-up; thyroglobulin (Tg); thyroid.

Introduction

Thyroglobulin (Tg) measurement following thyrotropin (TSH) stimulation is a useful tool in the management of patients affected by differentiated thyroid carcinoma (DTC) treated by thyroid ablation (i.e., thyroidectomy and radioiodine) (1, 2). Recently, Tg measurement during thyroxine (T4) treatment using high-sensitive assays (onT4-Tg) proved to be effective, even without TSH stimulation (3, 4). Using a high-sensitive immunoradiometric Tg assay, we previously found a 96% negative predictive value among patients undergoing T4 treatment. A further increase to 99% was obtained by coupling onT4-Tg and neck ultrasonography (US) (5). Globally, as confirmed even by using different high-sensitive Tg assays, few patients with undetectable onT4-Tg had a pathological Tg response (i.e., >2 ng/mL) to recombinant human TSH (rhTSH) stimulation, and less actually recurred (6, 7). Otherwise, no prospective data are available (8). The present study was undertaken to evaluate the impact of onT4-Tg measurement on both early and long-term outcome of patients with DTC.

Materials and methods

Patients

We enrolled 231 of 288 patients with histologically proven DTC [mean (SD) age, 52 (18) years; 24% male] papillary: 197, follicular: 34 with a low-risk profile according to European Thyroid Association Guidelines (2). Excluded were patients with 1. aggressive histotypes (i.e., papillary: tall-cell, columnar-cell, diffuse sclerosing; follicular: Hurtle-cell, widely invasive or poorly differentiated) (n=9); 2. maximum tumor diameter more than 40 mm and/or lymph-node(s) involve

*Corresponding author: PD Dr. med. Luca Giovanella, Nuclear Medicine and Thyroid Center, Oncology Institute of Southern Switzerland, 6500 Bellinzona, Switzerland
Phone: +41-91-8118672, Fax: +41-91-8118250,
E-mail: luca.giovanella@eoc.ch
Received November 27, 2008; accepted April 29, 2009; previously published online July 10, 2009

ment (N1) (n=39); 3. distant metastases (M1) (n=9). All patients were treated with total thyroidectomy and ^{131}I-iodide (3700 MBq) administration. Those with positive anti-thyroglobulin antibodies (TgAb) (n=33) and/or undetectable Tg before radioiodine ablation (n=3) were also excluded. After radioiodine ablation T4 was administered to suppress TSH (target <0.1 mUI/L with normal fT3) and the first follow-up visit 6 months later was characterized by neck US and serum TSH, Tg and TgAb measurements. Patients having detectable onT4-Tg (i.e., ≥0.2 ng/mL, see below) and/or positive US were further evaluated with US-guided fine-needle cytology (FNC) and/or other imaging procedures [i.e., ^{18}FDG-positron emission tomography/computed tomography (PET/CT), magnetic resonance imaging (MRI), PT-WBS]. If multi-imaging procedures tested negative, onT4-Tg was measured every 3 months and the Tg trend evaluated. Recurrences were confirmed by cytology, histology or specific radioiodine uptake. Patients with undetectable onT4-Tg and negative US were considered cured, T4 titred to obtain a TSH level from 0.1 mUI/L to 0.5 mUI/L and further visits, with neck US and TSH, Tg and TgAb measurements, performed every 12 months over an average of 6.8 (4.7–8.9) years of follow-up. Patients had ''no evidence of disease'' if no lesions were detected during follow-up (or Tg spontaneously normalized without treatment). Patients had ''disease recurrence'' when recurrences were proven histologically or lesions disappeared after specific radioiodine treatment. Finally, patients had ''biochemical recurrence'' if Tg increased progressively without any detectable lesion in multi-imaging procedures [including post-treatment whole body scan (PT-WBS)].

Methods

Tg measurement Serum Tg was assayed in duplicate using a specific IRMA assay (DYNOtest® Tg-plus, BRAHMS Diagnostica GmbH, Berlin, Germany) according to the manufacturer's instructions and our previously published reports (5, 9). Standardization of the assay is based on the international standard certified reference material (CRM) 457. One ng in the DYNOtest® Tg-plus is equivalent to 2 ng of CRM. Quality control consisted of measurement of two levels of control sera in each series. We reanalyzed showing a coefficient of variation exceeding 10% and participated bimonthly in the European interlaboratory control Oncocheck. This method showed a functional sensitivity of 0.2 ng/mL; consequently serum Tg concentrations <0.2 ng/mL were reported as undetectable (9, 10).

Screening for interferences The presence of anti-thyroglobulin antibodies (TgAb) was assessed using a specific radioimmunoassay (DYNOtest® anti-TGn, BRAHMS Diagnostica GmbH, Berlin, Germany) and a recovery test performed using buffer provided by the manufacturer with a cut-off value of 80%. Samples with TgAb>60 UI/mL and/or recovery <80% were excluded. All samples were retested after incubation in heterophile-blocking tubes (HBT; Scantibodies, Santee, CA, USA) at room temperature for 1 h. Samples with Tg falling to <30% of their original value, becoming undetectable or increasing more than 30% of their original value were considered as having interference and excluded.

TSH and fT3 assay Serum TSH and fT3 concentrations were measured using immunochemiluminometric assays on the Immulite 2000 platform (Diagnostic Products Corporation, Los Angeles, CA, USA).

Results

As shown in Figure 1, seven of 195 (3.5%) patients had detectable onT4-Tg (from 0.3 ng/mL to 1.4 ng/mL) with a corresponding suppressed TSH (i.e., <0.1 mUI/L) at the first follow-up visit; two had neck lymph node metastases (onT4-Tg 0.6 and 1.1 ng/mL, respectively), one recurred in the thyroid bed (onT4-Tg 0.5 ng/mL) and one in a mediastinal lymph node (onT4-Tg 1.4 ng/mL). Lesions were histologically confirmed (n=3) or resolved after radioiodine treatment (n=1). A spontaneous reduction in onT4-Tg from 0.3, 0.5 and 0.8 ng/mL to undetectable concentrations occurred in three patients over a 3–6 months period; none of these relapsed in 3.7–7.1 years of follow-up. Two of 188 (1%) patients with undetectable onT4-Tg at first visit showed recurrence during long-term follow-up. In the first patient, serum onT4-Tg became detectable 9 months following thyroid ablation (0.3 ng/mL). Five months later two neck lymph node

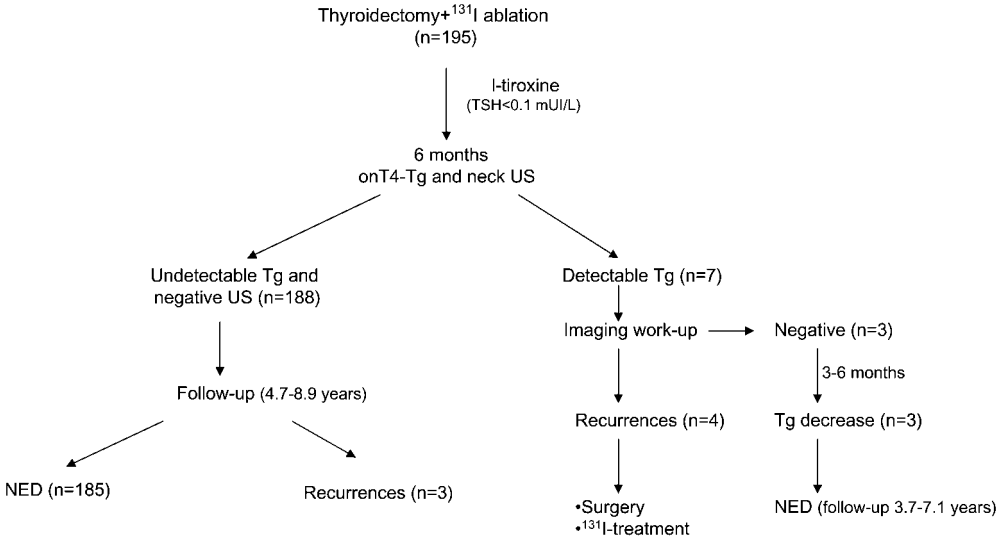

Figure 1 Follow-up and outcome of 195 patients with low-risk of DTC.

metastases (6 and 8 mm) were detected by US with a corresponding onT4-Tg increase to 0.9 ng/mL. The second patient showed an increase in onT4-Tg (0.4 ng/mL) 15 months following thyroid ablation. Seven months later, neck US revealed an 11 mm lymph node recurrence with a corresponding onT4-Tg of 1.3 ng/mL. Recurring tumors were resected, and in both cases Tg became undetectable (even after rhTSH stimulation) with negative follow-up at 3.4 and 2.9 years, respectively (Figure 2). Finally, a progressive increase in onT4-Tg from <0.2 ng/mL to 1.3 ng/mL in a 32-month period was observed in a patient (0.5%) with negative results in multiple imaging procedures (i.e., biochemical recurrence). None of the remaining 185 patients (98.5%) showed recurrence over 6.8 (4.7–8.8) years of follow-up. Twelve of these (6.4%) exhibited a slight increase in serum onT4-Tg (ranging from 0.2 ng/mL to 0.6 ng/mL) that spontaneously fell to undetectable levels within 3–6 months (i.e., negative Tg-trend).

Discussion

European and American Thyroid Association Guidelines suggest that a patient with DTC can be considered free of disease when there is no clinical evidence of tumor, no imaging evidence of tumor and the serum Tg is undetectable during TSH suppression therapy and following TSH stimulation (1, 2). However, some authors found a negligible role for TSH stimulation in a large number of patients having DTC, primarily those with a low-risk profile (4–6, 11, 12). Accordingly, we selected DTC patients with a low-risk profile according to the European Thyroid Association Guidelines, and evaluated the impact of serum onT4-Tg measurement in both early and long-term outcome while TSH stimulation was omitted. Six months following thyroid ablation, an increase in serum onT4-Tg occurred in four and three patients with ($n=4$) and without ($n=191$) proven DTC recurrence, respectively (sensitivity 100%, specificity 98%, positive predictive value 98%, negative predictive value 100%, accuracy 98%). Our data agree with those of Schlumberger and co-workers who showed that Tg immunoassays, with a functional sensitivity of 0.2–0.3 ng/mL (comprising the assay employed in our study), are accurate enough to rule out TSH stimulation in the first follow-up visit of 944 patients with DTC (13). Here, we also show that false positive results were always associated with a spontaneous decrease within 3–6 months, proving that a negative Tg trend has 100% negative predictive value. Three of 188 patients negative during early follow-up showed recurrence with a progressive increase of onT4-Tg in all cases. These results agree with data from Zophel and co-workers who studied 126 patients with radically cured disease over a 4-year period. The Tg levels increased in five (4%) patients; proven recurrence was seen in four patients while the fifth remained well, despite rising serum Tg concentrations that could be stimulated by TSH (14). Finally, a transient increase in onT4-Tg occurred in 12 disease-free patients during long-term follow-up. However, the marker spontaneously declined within 3–6 months in all cases. As a result, monitoring the onT4-Tg trend allowed us to accurately discriminate between patients with recurrence of DTC from those without recurrences.

Limitations

DTC recurrence can occur decades following thyroid ablation. Consequently, life-long monitoring is required. In this respect, the follow-up period needs to be longer for our patients, and our data should be regarded as preliminary. However, most recurrent or persistent tumor is detected during the first 5 years following thyroidectomy (15). Our patients will be monitored further in order to evaluate overall survival and cancer related morbidity over a longer period.

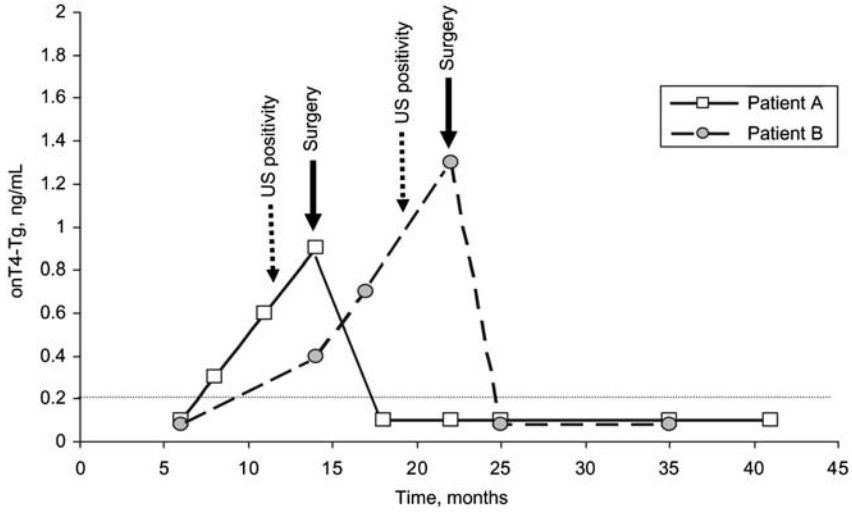

Figure 2 Trend of serum onT4-Tg in two patients with recurrent DTC during long-term follow-up. The dashed line represents the functional sensitivity of the assay.

Conclusions

Undetectable serum onT4-Tg with a high-sensitive immunoradiometric assay following thyroid ablation predicts low-risk of DTC recurrence. When onT4-Tg became detectable during follow-up, the evaluation of the Tg-slope in a 3–6 month period accurately discriminated between patients recurrence of DTC from those without recurrence. This helps to avoid unnecessary diagnostic or therapeutic procedures.

References

1. Mazzaferri E, Robbins RJ, Spencer CA, Bravermann LE, Pacini F, Wartofsky L, et al. A consensus report of the role of serum thyroglobulin as a monitoring method for low-risk patients with papillary thyroid carcinoma. J Clin Endocrinol Metab 2003;88:1433–41.
2. Pacini F, Schlumberger M, Dralle H, Elisei R, Smit JW, Wiersinga W, et al. European consensus for the management of patients with differentiated thyroid carcinoma of the follicular epithelium. Eur J Endocrinol 2006;154:787–803.
3. Souza do Rosario PW, Ribeiro Borges MA, Fagundes TA, Franco AC, Purisch S. Is stimulation of thyroglobulin (Tg) useful in low-risk patients with thyroid carcinoma and undetectable Tg on thyroxin and negative neck ultrasound? Clin Endocrinol 2005;62:121–5.
4. Persoon AC, Jager PL, Sluiter WJ, Plukker JT, Wolffenbuttel BH, Links TP. A sensitive Tg assay or rhTSH stimulated Tg: what's the best in the long-term follow-up of patients with differentiated thyroid carcinoma? PLoS ONE 2007;2:e816. Accessed Oct 6, 2008.
5. Giovanella L, Ceriani L, Ghelfo A, Maffioli M, Keller F, Spriano G. Thyroglobulin assay during thyroxine treatment in low-risk differentiated thyroid cancer management: comparison with recombinant thyrotropin-stimulated assay and imaging procedures. Clin Chem Lab Med 2006;44:648–52.
6. Smallridge RC, Meek S, Morgan MA, Gates GS, Fox TP, Grabe S, et al. Monitoring thyroglobulin in a sensitive immunoassay has comparable sensitivity to recombinant human TSH-stimulated thyroglobulin in follow-up of thyroid cancer patients. J Clin Endocrinol Metab 2007;92:82–7.
7. Iervasi A, Iervasi G, Ferdeghini M, Solimeo C, Bottoni A, Rossi L, et al. Clinical relevance of highly sensitive Tg assay in monitoring patients treated for differentiated thyroid cancer. Clin Endocrinol 2007;67:434–41.
8. Mazzaferri EL. Will highly sensitive thyroglobulin assays change the management of thyroid cancer? Clin Endocrinol 2007;67:321–3.
9. Giovanella L, Ceriani L. High-sensitivity human thyroglobulin (hTG) immunoradiometric assay in the follow-up of patients with differentiated thyroid cancer. Clin Chem Lab Med 2002;40:480–4.
10. Morgenthaler NG, Froelich J, Rendl J, Willnich M, Alonso C, Bergmann A, et al. Technical evaluation of a new immunoradiometric and a new immunoluminometric assay for thyroglobulin. Clin Chem 2002;48:1077–83.
11. Rosario PW, Purisch S. Does a highly sensitive thyroglobulin (Tg) assay change the clinical management of low-risk patients with thyroid cancer with Tg on T4 $<$ 1 ng/mL determined by traditional assays? Clin Endocrinol 2008;68:338–42.
12. Giovanella L. Highly sensitive thyroglobulin measurements in differentiated thyroid carcinoma management. Clin Chem Lab Med 2008;46:1067–73.
13. Schlumberger M, Hitzel A, Toubert ME, Corone C, Troalen F, Schlageter MH, et al. Comparison of seven serum thyroglobulin assays in the follow-up of papillary and follicular thyroid cancer patients. J Clin Endocrinol Metab 2007;92:2487–95.
14. Zophel K, Wunderlich G, Smith BR. Serum thyroglobulin measurements with high sensitivity enzyme-linked immunosorbent assay: is there a clinical benefit in patients with differentiated thyroid carcinoma? Thyroid 2003;13:861–5.
15. Mazzaferri EL, Shiang SM. Long-term impact of initial surgical and medical therapy on papillary and follicular thyroid carcinoma. Am J Med 1994;97:418–28.